Problems with estimating vitamin C intakes[1,2]

Rashmi Sinha, Gladys Block, and Philip R Taylor

ABSTRACT The vitamin C content of foods was examined from two national databases and new values were obtained by HPLC. HPLC values were lower in four of the five highest vitamin C contributors to the US diet (orange juice, grapefruit, tomatoes and tomato juice, and potatoes), as well as in broccoli, red peppers, and cooked collard and mustard greens, compared with values from the other databases. When HPLC values were substituted in the Health Habits and History Questionnaire, the resulting estimates of dietary intake of vitamin C in two studies were lower. Despite these lower estimates of absolute intake, in one study the correlation between dietary vitamin C and plasma ascorbic acid was similar. In conclusion, the accuracy of the vitamin C content of foods is important for estimating the absolute amount of vitamin C intake in the population but may not change the ranking of people in epidemiological studies. *Am J Clin Nutr* 1993;57:547–50.

KEY WORDS Vitamin C, ascorbic acid, dehydroascorbic acid, National Health Interview Survey, National Health and Nutrition Examination Survey, high-performance liquid chromatography

Introduction

Problems exist in estimating both long- and short-term dietary intakes of nutrients. Variability within individual's intake and recall errors have received considerable attention, but nutrient-composition data can also contribute to errors in estimating nutrient intake. Nutrient compositional data errors may reflect inaccurate nutrient values in foods if there are different magnitudes or direction (above or below the actual values) of errors in estimating nutrients in foods. Moreover, if foods vary in their importance by subgroups, such nutrient composition errors can lead to differential misclassification and also to errors in estimating subgroup intake. This paper investigates the impact of nutrient-composition data on population estimates of vitamin C intake. Similar problems may be true for other nutrients, such as vitamins A, B-12, D, and E; folacin; pantothenic acid; and carotenes. Stewart (1) noted that the probability of a correct analytical value for these nutrients was only "fair."

Newer methods for nutrient analysis that may provide both more accurate and precise measurement are continually being developed. Current state-of-the-art analysis for ascorbic acid (AA) is HPLC. Vanderslice and Higgs (2–7) used HPLC in conjunction with robotic extraction to determine both AA and dehydroascorbic acid (DHAA) contents of selected foods known to provide most of the vitamin C in the normal diets consumed in the United States (8). Use of the HPLC values may provide a more accurate estimate of vitamin C intake from dietary questionnaires and thus provide better estimates of the role of diet in health and disease.

In this paper we compare the vitamin C content of foods in two existing nutrient-composition databases with values from recent HPLC analyses. We then compare the nutrient-intake estimates from the old and new nutrient-composition data in two surveys, using a food frequency questionnaire.

Methods

The AA and DHAA contents of major contributors of vitamin C in the US diet were analyzed by Vanderslice et al (7). Food was purchased in the Washington, DC area at local supermarkets between November 1988 and May 1989. Sample and market variability in the vitamin C content of banana, snap beans, broccoli, cabbage, grapefruit, orange, potato, spinach, and tomato were also examined. Foods were prepared by using *Better Homes & Gardens New Cookbook* (9).

The HPLC values were then compared with AA content given by the US Department of Agriculture's (USDA) revised *Handbook no. 8* (10) for comparable foods and those given by the database for the Health Habits and History Questionnaire (HHHQ) (11). The HHHQ was developed by using dietary-intake data from the second National Health and Nutrition Examination Survey (NHANES II) (8, 12). The 2244 different food codes from the original NHANES II responses were grouped into 147 conceptually similar food items. Foods were included in the HHHQ if they made an important contribution to the population's intake of energy and each of 17 nutrients in the NHANES II database. The quantitative nutrient values assigned for each food on the questionnaire were also developed in a data-based manner, which made use of the frequency and portion-size information from the NHANES II survey. The nutrient values in the NHANES II database were taken from USDA data tapes 456-1 and 456-2 and from manufacturers for commercial foods that were reported ≥ 20 times (CM Dresser, from Proceedings of the Eighth National Nutrient Data Base Conference, 1983). Most vitamin C values from NHANES II and USDA *Handbook no. 8* were obtained by colorimetry.

[1] From the Division of Cancer Prevention and Control, National Cancer Institute, Bethesda, MD, and the Department of Public Health Nutrition, University of California, Berkeley.

[2] Address reprint requests to R Sinha, EPN Room 443, National Cancer Institute, 9000 Rockville Pike, Bethesda, MD 20892.

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TABLE 1
Vitamin C contents of selected food items in forms most frequently eaten*

Food items from HHHQ	Original HHHQ† (reduced AA)	USDA handbook no. 8 (reduced AA)	HPLC analysis‡		Total AA
			Reduced AA§	DHAA	
	mg/g wet wt (mg/100 g wet wt)				
Vegetables					
String bean, green beans (fresh boiled)	0.04 (4.0)	0.10 (9.7)	0.07 (6.7)	0.01 (1.3)	0.08 (8.0)
Tomatoes, tomato juice (fresh raw)	0.23 (23.0)	0.18 (18.0)	0.11 (10.6)	0.03 (3.0)	0.14 (13.6)
Broccoli (fresh boiled)	0.90 (90.0)	0.63 (62.8)	0.37 (37.0)	0.03 (2.6)	0.40 (39.6)
Cauliflower or brussel sprouts (fresh raw)‖	0.55 (55.0)	0.72 (71.5)	0.54 (54.0)	0.09 (8.7)	0.63 (62.7)
Spinach (raw)	0.51 (51.0)	0.28 (28.1)	0.52 (52.4)	—	0.52 (52.4)
Spinach (cooked)	0.19 (19.0)	0.10 (9.8)	0.15 (15.1)	—	0.15 (15.1)
Mustard greens, turnip greens, collards (boiled for 1 h)	0.48 (48.0)	0.25 (25.3)	0.05 (4.8)	—	0.05 (4.8)
Coleslaw (1-day old), cabbage, sauerkraut	0.20 (29.0)	0.33 (32.7)	0.21 (20.7)	0.10 (9.6)	0.30 (30.3)
Carrots or mixed vegetables with carrots (fresh raw)	0.08 (8.0)	0.09 (9.3)	0.05 (4.7)	0.02 (2.0)	0.07 (6.7)
Mixed green salad	0.11 (10.6)	—	0.08 (7.9)	0.03 (2.8)	0.11 (10.7)
Sweet red peppers	2.04 (204.0)	1.28 (128.0)	1.51 (151.0)	0.04 (4.0)	1.55 (155.0)
French fries (fast food)	0.21 (21.0)	0.10 (10.3)	0.14 (14.3)	0.09 (9.0)	0.23 (23.3)
Other potatoes, including boiled, baked, or potato salad (boiled without skin)	0.12 (12.3)	0.11 (10.7)	0.07 (7.0)	0.01 (1.3)	0.08 (8.3)
Fruits					
Bananas	0.10 (10.0)	0.09 (9.1)	0.15 (15.3)	0.03 (3.3)	0.19 (18.6)
Cantaloupe	0.33 (33.0)	0.42 (42.2)	0.28 (28.0)	0.03 (2.7)	0.31 (30.7)
Grapefruit	0.37 (37.0)	0.33 (33.3)	0.21 (21.3)	0.02 (2.3)	0.24 (23.6)
Orange	0.50 (50.0)	0.52 (52.2)	0.59 (58.7)	0.60 (5.6)	0.64 (64.3)
Watermelon	0.07 (7.0)	0.10 (9.6)	0.08 (8.0)	0.02 (1.7)	0.10 (9.7)
Juices					
Orange juice or grapefruit juice (1-d old, reconstituted)	0.48 (48.0)	0.36 (36.3)	0.33 (33.0)	0.04 (4.3)	0.37 (37.3)
Tang, Start breakfast drinks	0.35 (35.0)	—	0.57 (57.3)	0.02 (2.0)	0.59 (59.3)
Other foods					
Pizza (fast food)	0.06 (6.0)	0.02 (2.0)	0.09 (0.9)	0.003 (0.3)	0.01 (1.2)
Spaghetti (with tomato sauce), lasagna, other pasta	0.09 (9.0)	—	0.01 (1.0)	0.005 (0.5)	0.02 (1.5)
Highly fortified cereals (without milk; eg, Product 19)¶	2.12 (212.0)	2.12 (212.0)	2.26 (226.0)	0.38 (38.0)	2.64 (264.0)
Other cold cereals (without milk) (eg, Corn Flakes)	0.53 (53.0)	0.53 (53.0)	0.60 (59.7)	0.03 (2.5)	0.62 (62.2)
Hot dogs	0.0**	0.001 (0.1)††	0.18 (18.0)	0.03 (3.0)††	0.21 (21.0)

* HHHQ, Health Habits and History Questionnaire; USDA, US Department of Agriculture; AA, ascorbic acid; DHAA, dehydroascorbic acid. Where no values are listed, the concentration was < 0.01 mg/g sample.

† Reduced vitamin C values used in HHHQ were derived by using the National Health and Nutrition Examination Survey (NHANES II) nutrient database.

‡ Analyzed by Vanderslice et al (7).

§ For other vitamin C containing foods on the HHHQ questionnaire, which are relatively unimportant sources of this vitamin, the existing estimates may be used.

‖ The value of vitamin C for "cauliflower or brussel sprouts" is for cauliflower because it is the predominant food in that group.

¶ Kellogg, Battle Creek, MI.

** NHANES II gives a vitamin C value for hot dogs but HHHQ does not, because in some products isoascorbic acid rather than AA is added.

†† Hot dog analyzed in USDA Handbook no. 8 is with a bun whereas the HPLC analysis is for hot dog without a bun.

To further analyze the impact of nutrient-content differences on estimates of dietary intakes, we examined intake data from three studies, NHANES II (8), the 1987 National Health Interview Survey (NHIS) (13), and a vitamin C study in humans conducted by the National Cancer Institute (NCI) in collaboration with the USDA (NCI/USDA) (14, 15).

NHANES II included a large representative sample of the US population and was conducted between 1976 and 1980 by the National Center for Health Statistics (13). Dietary data were collected by using 24-h recalls, and serum AA was analyzed by the Centers for Disease Control, Atlanta (16) by using the 2,4-dinitrophenylhydrazine method (17).

Dietary intakes for the NHIS and the NCI/USDA study were both obtained by using the HHHQ food frequency questionnaire (11). The HHHQ provides estimates of usual dietary intake of energy and macro- and micronutrients and has been the subject of several validation studies (18–20).

The 1987 NHIS used a 60-food item version of the HHHQ to obtain data on all adults aged ≥ 18 y within a household, a total of 22 080 persons (21, 22). In the NCI/USDA study, 68

TABLE 2
Dietary intake of vitamin C and plasma ascorbic acid (AA) in various studies*

	Estimated dietary vitamin C†		Plasma total AA
	Old value	New value	
	mg	*mg*	*μmol/L*
NHIS‡			
Male			
White	125.0	93.2	—
Black	144.1	106.9	—
Female			
White	112.0	82.3	—
Black	127.5	93.6	—
NCI/USDA Study†§			
Non-vitamin C Supplement user	124.6 (r = 0.50)‖	114.6 (r = 0.48)	60
NHANES II¶			
Male			
White	106.3	—	50
Black	110.0	—	40
Female			
White	94.6	—	70
Black	99.8	—	50

* NHIS, National Health Interview Survey; NCI, National Cancer Institute; USDA, US Department of Agriculture; NHANES II, Second National Health and Nutrition Examination Survey.

† Estimates are for reduced vitamin C (10).

‡ In both the NHIS and NCI/USDA studies the diet estimates are based on the Health Habits and History Questionnaire.

§ The vitamin C study was carried out in 30–59-y-old healthy males.

‖ Correlation between dietary reduced AA and plasma reduced AA.

¶ Vitamin C intake values from the NHIS and NCI/USDA studies may not be directly comparable with those from NHANES II because of the different methods used to estimate intakes.

healthy males (aged 30–59 y; 52 whites, 8 blacks, 5 Hispanic, and 3 Asian) completed a 100-food item HHHQ. At the same time, fasting blood samples were collected and plasma was analyzed for AA by using the 2,4-dinitrophenylhydrazine assay (17).

Vitamin C intake in the NHIS survey and NCI/USDA study were first estimated by using the original database values of the HHHQ, which were based on the NHANES II. Nutrient-composition values for vitamin C were then replaced by the HPLC values of Vanderslice et al (7) in the HHHQ database, and dietary vitamin C intakes in the NHIS and NCI/USDA studies were reestimated. Finally, in the NCI/USDA study, we calculated Pearson correlation coefficients between plasma AA and dietary AA as estimated by the HHHQ, using original and newer HPLC values (23).

Results and Discussion

Table 1 gives reduced and dehydro forms of ascorbate for the major foods sources of vitamin C in the HHHQ database. There are three values of vitamin C: those used in the original HHHQ database (based on the NHANES II data), those found for comparable foods in the revised USDA *Handbook no. 8* published in 1988 (10), and the HPLC measurements of vitamin C in selected food items (7). The NHANES II and USDA *Handbook no. 8* values are purported to represent primarily reduced rather than total AA (AA + DHAA), but in some instances they may be based on studies in which the distinction was not made.

There are differences between estimates of the vitamin C content of foods when the three databases are examined. Vitamin C values for the major contributors of this vitamin in the American diet (24)—orange juice (26.5%), grapefruit (7.2%), tomatoes and tomato juice (6.1%), and potatoes (4.2%)—are all lower in the USDA *Handbook no. 8* printed in 1988 than in the NHANES II database. The newer HPLC values of vitamin C in these foods are even lower.

Large discrepencies were observed in the vitamin C content of broccoli, red peppers, and cooked greens such as mustard, turnip greens, and collards (**Table 1**). These differences may be important if much of the vitamin C for individuals or subgroups comes from one of these foods. This is the case, for example, for cooked greens among blacks (8). Block and Sorenson (8) reported that greens contribute > 10% of dietary vitamin C for blacks. The vitamin C content of greens estimated by NHANES II is 0.48 mg/g wet wt (48.0 mg/100 g wet wt). However, when greens are boiled for 1 h the value decreases to 0.253 mg/g wet wt (25.3 mg/100 g wet wt) by USDA *Handbook no. 8* values, but even further to 0.048 mg/g wet wt (4.8 mg/100 g wet wt) when analyzed by HPLC. Therefore, greens as usually prepared and consumed may actually contribute very little vitamin C in the diets of blacks. The impact of this discrepency in estimating vitamin C intake is likely to be much higher for the black than for the white population.

Table 2 shows that the recalculated vitamin C intake estimates from the NHIS and the NCI/USDA studies show substantially lower values in all age and sex groups when the HPLC values are used. This suggests that vitamin C intake in the American population may actually be lower than what is assumed to be true based on the NHANES II database (shown in Table 2 for comparison) or any population estimates generated by using the USDA database. However, a direct inference in the magnitude of effect on vitamin C intake estimates cannot be made from NHIS and NCI/USDA studies to NHANES II. Two different dietary instruments were used in these studies and they contrast in accuracy and types of biases contained within them (25). NHIS and NCI/USDA studies used a food frequency questionnaire to estimate nutrient intakes whereas NHANES II used 24-h recalls.

When correlations between dietary and plasma AA in the NIH/USDA study were recalculated by using the HPLC (r = 0.48, $P < 0.0009$) vs the original HHHQ (r = 0.50, $P < 0.0004$) values, there was essentially no difference in the correlations. Thus, although the absolute values of the intake estimate were affected by using HPLC values, ranking was not, at least in the NCI/USDA study group.

This robustness of the correlation between diet and plasma AA despite differences in the dietary values suggests that the frequency of consumption and portion size of key foods is more important than small differences in the nutrient content of foods in determining plasma values. Therefore, in epidemiological studies, the updated vitamin C values of foods may not be crucial for purposes of ranking or classifying individuals with regard to vitamin C intake. However, we have shown that the new values do produce substantial differences in estimates of the absolute vitamin C intake in the population.

In summary, the new values for vitamin C presented here may provide a better estimation of vitamin C intake than the ones currently being used, which tend to overestimate intakes.

The HPLC values also afford investigators the ability to examine separately the effects of AA in its reduced and DHAA forms.

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